

Appl. No. 09/838,382
Amendment dated December 18, 2003
Reply to Office action of June 18, 2003

Amendments to the Specification:

Please replace the first sentence of the paragraph beginning at Page 11, Line 24, with the following rewritten sentence:

The *Eimeria* strain *Eimeria maxima*-I has been deposited with the American Type Culture Collection (ATCC), 10801 University Blvd., Manassas, Va. 20110-2209 on July 25, 2002 under accession number [[]] PTA-4959 as a patent [[deposits]] deposit under the Budapest Treaty on the International Recognition of the Deposit of Microorganisms for the Purposes of Patent Procedure.

Please replace the paragraph bridging Pages 1 and 2 with the following amended paragraph:

Vaccination against coccidiosis is not a new concept; it has been used by the poultry industry since the early 1950s. The four dominant commercial vaccines used by the poultry industry, [[Coccivac®, Immucox®]] COCCIVAC®, IMMUCOX®, Livacox, and [[Paracox®]] PARACOX®, typically incorporate several species and strains of *Eimeria*. Some of the species in the vaccines are attenuated, egg-adapted and/or precocious lines, while others are strains that were originally isolated from commercial poultry production facilities. While all of these vaccines provide solid immunity to coccidial infection when applied carefully under good rearing conditions, especially in replacement and breeding flocks, these vaccines have not been universally accepted by the U.S. poultry industry for meat-producing broiler and heavy roaster bird flocks. The two major reasons for this reluctance has been (1) that bird performance, as measured by weight gain and feed efficiency, has not always equaled that seen with prophylactically medicated birds and (2) that reports of immunological variability of *E. maxima*, one of the major pathogenic and economically important coccidial species,

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indicate that vaccination with a given suspension of live oocysts may not be effective in protecting against field strains in differing geographical locations (Long *et al.* 1986. *Avian Pathol.* 15: 217-278; Long *et al.* 1979. *Parasit.* 79: 451-457; Rose, M.E. 1982. In: The Biology of the Coccidia, P. L. Long, Ed., University Park Press, Baltimore MD, pages 329-371; Fitz-Coy, S. H. 1992. *Avian Dis.* 36: 40-43; Lee, E.-H. 1993. In: Proceedings of the VIth International Coccidiosis Conference, Barta and Fernando, Eds., University of Guelph, Guelph, Ont., Canada, pages 118-121).

Please replace the paragraph of Example 6, Page 17 with the following amended paragraph:

For this trial, groups of approximately 50 chicks were each inoculated at 1 day of age, as described in Example 2. Birds were first immunized with the *E. maxima* -GPL strain, the immunovariant strain, *E. maxima* -I, or a third *E. maxima* field strain (designated *E. maxima* -ESS) and then challenged with each of these three strains. Male birds were immunized with 100 oocysts/ bird at 1 day of age or 1000 oocysts/ bird at 10 days of age given by oral gavage and challenged with 25,000 oocysts of *E. maxima* -I/ bird, 30,000 oocysts of *E. maxima* -GPL/ bird, or 50,000 oocysts of *E. maxima* -ESS/ bird at 20 days of age. In addition, birds immunized with a commercially available live oocyst vaccine [[Coccivac B®]] COCCIVAC B® (American Scientific Laboratories, Millsboro, DE; 0.1ml of 10 dose/ml) or the [[Coccivac B®]] COCCIVAC B® vaccine (10 dose/ml) to which the *E. maxima*-I strain (1000 oocysts/ml; 0.1 ml/bird) was added, were also challenged with the three different *E. maxima* strains. Parameters measured were intestinal lesion scores, average weight gain during challenge infection, average weight gain from boost to termination of the experiment, and feed conversions (amount of feed consumed/weight gain). Table 2 shows the result for the parameters listed above after

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challenge of birds immunized with *E. maxima* -GPL, *E. maxima* -I, *E. maxima* -ESS, [[Coccivac B®]] COCCIVAC B®, or [[Coccivac B® -*E. maxima* -I]] COCCIVAC B® - *E. maxima* -I with the three different strains of *E. maxima*.

Please replace Table 2, Pages 18 and 19 with the following amended Table 2:

Table 2. Caged Battery Trial Experiment Using Male Broiler Birds

<u>Treatment</u> Immunize / Challenge	Avg Feed Conver.	Avg Lesion Score During Challenge*	Avg Wgt Gain During Challenge*	Avg Wgt Gain - End Challenge*
Non/Non	1.74	0.00 ^a	373.47 ^a	903.04 ^a
Non/ <i>E. maxima</i> -I	3.23	3.77 ^b	158.90 ^c	683.42 ^c
Non/ <i>E. maxima</i> -ESS	1.96	3.31 ^b	294.44 ^b	816.31 ^b
Non/ <i>E. maxima</i> -GPL	3.11	3.61 ^b	153.13 ^c	675.66 ^a
[[Coccivac®- <i>E. maxima</i> -I/ Non]] <u>COCCIVAC®-<i>E. maxima</i> -I/ Non</u>	1.75	0.08 ^a	361.61 ^a	860.64 ^a
[[Coccivac®- <i>E. maxima</i> -I/]] <u>COCCIVAC®-<i>E. maxima</i> -I/</u> <i>E. maxima</i> -GPL	1.80	0.69 ^b	335.25 ^a	861.56 ^a
[[Coccivac®- <i>E. maxima</i> -I/]] <u>COCCIVAC®-<i>E. maxima</i> -I/</u> <i>E. maxima</i> -I	1.81	0.00 ^a	342.43 ^a	860.12 ^a
[[Coccivac®- <i>E. maxima</i> -I/]] <u>COCCIVAC®-<i>E. maxima</i> -I/</u> <i>E. maxima</i> -ESS	1.77	0.00 ^a	365.00 ^a	882.14 ^a

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<u>Treatment</u>	Avg Feed Conver.	Avg Lesion Score During Challenge*	Avg Wgt Gain During Challenge*	Avg Wgt Gain - End Challenge*
<i>E. maxima</i> -I/ Non	1.69	0.07 ^a	388.93 ^a	912.86 ^a
<i>E. maxima</i> -I/ <i>E. maxima</i> -GPL	1.86	1.37 ^b	327.74 ^b	833.64 ^b
<i>E. maxima</i> -I/ <i>E. maxima</i> -I	1.68	0.00 ^a	371.69 ^{ab}	884.69 ^{ab}
<i>E. maxima</i> -I/ <i>E. maxima</i> -ESS	1.81	0.00 ^a	347.48 ^{ab}	873.00 ^{ab}
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[[Coccivac®/Non]]	1.71	0.00 ^a	395.57 ^a	934.14 ^a
<u>COCCIVAC®/Non</u>				
[[Coccivac®/ <i>E. maxima</i> -GPL]]	1.85	2.13 ^b	313.67 ^b	797.94 ^c
<u>COCCIVAC®/ <i>E. maxima</i>-GPL</u>				
[[Coccivac®/ <i>E. maxima</i> -I]]	2.36	3.21 ^c	224.63 ^c	740.13 ^c
<u>COCCIVAC®/ <i>E. maxima</i>-I</u>				
[[Coccivac®/ <i>E. maxima</i> -ESS]]	1.91	2.08 ^b	303.50 ^b	826.31 ^b
<u>COCCIVAC®/ <i>E. maxima</i>-ESS</u>				
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<i>E. maxima</i> -GPL/ Non	1.79	0.00 ^a	367.42 ^a	888.90 ^{ab}
<i>E. maxima</i> -GPL/ <i>E. maxima</i> -GPL	1.70	0.06 ^a	365.19 ^a	904.56 ^a
<i>E. maxima</i> -GPL/ <i>E. maxima</i> -I	1.92	2.25 ^b	306.88 ^b	828.19 ^b
<i>E. maxima</i> -GPL/ <i>E. maxima</i> -ESS	1.79	0.64 ^a	334.36 ^{ab}	836.36 ^{ab}
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<i>E. maxima</i> -ESS/ Non	1.72	0.06 ^a	387.79 ^a	956.67 ^a
<i>E. maxima</i> -ESS/ <i>E. maxima</i> -GPL	3.05	3.93 ^c	166.39 ^c	711.63 ^c
<i>E. maxima</i> -ESS/ <i>E. maxima</i> -I	3.22	3.63 ^c	168.31 ^c	715.56 ^c
<i>E. maxima</i> -ESS/ <i>E. maxima</i> -ESS	1.88	2.63 ^b	313.88 ^b	861.94 ^b

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Please replace the last paragraph on Page 19 with the following amended paragraph:

Based on the statistical analysis of these parameters, *E. maxima* -GPL immunization did not protect against *E. maxima*-I challenge, *E. maxima*-I immunization did not protect against *E. maxima*-GPL challenge, and *E. maxima*-ESS immunization did not protect against *E. maxima* -GPL, *E. maxima*-I, or even its homologous challenge with *E. maxima*-ESS. Birds immunized with ~~[[Coccivac B®]]~~ COCCIVAC B® vaccine were not protected against challenge by any of the three *E. maxima* strains even though the vaccine did contain its own strain of *E. maxima*. However, the addition of the *E. maxima*-I strain to the ~~[[Coccivac B®]]~~ COCCIVAC B® vaccine did significantly protect the immunized birds against challenge with the three different *E. maxima* strains. Addition of the *E. maxima*-I strain to the live oocyst vaccine increased the effectiveness of the vaccine against immunovariant strains of *E. maxima* that could be present in broiler grow-out houses by eliciting a more uniform immunological protection against coccidial infection.